

## RECEIVED

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<110> Dahlqvist, Andres Stahl, Ulf Lenman, Marit Banas, Antoni Ronne, Hans

<120> A new class of enzymes in the biosynthetic pathway for the production of triacylglycerol and recombinant DNA molecules encoding these enzymes

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Rubi DI

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Ala His Val Asp Ile Met Gly Asn Phe Ala Leu Tle Glu Asp Ile Met 625 630 635 \ 640

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Thr Gln Ser Gly Ala His Val\Asp Ile Met Gly Asn Phe Ala Leu Ile
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Arg Lys Ser Lys Phe Gly Lys Arg Leu Ash Phe Ile Leu Gly Ala Ile
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Leu Gly Ile Cys Gly Ala Phe Phe Phe Ala Val Gly Asp Asp Asn Ala 65 70 80

Val Phe Asp Pro Ala Thr Leu Asp Lys Phe Gly Asn Met Leu Gly Ser 85 90 95

Ser Asp Leu Phe Asp Asp Ile Lys Gly Tyr Leu Ser Tyr Asn Val Phe
100 105 110

Lys Asp Ala Pro Phe Thr Thr Asp Lys Pro Ser Gly
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Trp Leu Glu His Leu Met Leu Asp Lys Lys Thr Gly Leu Asp Pro Lys

Gly Ile Lys Leu Arg Ala Ala Gln Gly Phe Glu Ala Ala Asp the Phe

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Ile Gly Tyr Glu Pro Asn Asn Met Leu Ser Ala Ser Tyr Asp Trp Arg
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Pro Lys Thr Val Ala Ala Leu Leu Ser Gly Glu Met Lys Asp Thr Gly

345

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Lys Gly Gly Asp Val Ala Pro Asp Asp Leu Asn Gln Thr Asn Phe Ser 385 390 400

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Glu Phe Asp Ile Asp Asp Ala Leu Gln Phe Leu Lys Asn Val Thr Asp
420 425 430

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His Val Asp Ile Leu Gly H\s Ser Glu Leu Asn Glu Ile Ile Leu Lys
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Tyr Pro Ile His Lys Lys Ser Gly Gly Trp Phe Arg Leu Trp Phe Asp
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Ala Ala Val Leu Leu Ser Pro Phe Thr Arg Cys Phe Ser Asp Arg Met
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Met Leu Tyr Tyr Asp Pro Asp Leu Asp Asp Tyr dln Asn Ala Pro Gly
100 105 110

Val Gln Thr Arg Val Pro His Phe Gly Ser Thr Lys Ser Leu Leu Tyr
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Leu Lys Val Asp Ser Leu Asn Thr Val Glu Ile Asp Gly Val Ser His 385 390 395 400

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Lys Arg Asp Gly Asn Gly Arg Lys Arg Trp Arg Asp Ser Arg Arg Leu
65 70 75 80

Ile Phe Ile Leu Gly Ala Phe Leu Gly Val Leu Leu Pro Phe Ser Phe 85 Gly Ala Tyr His Val His Asn\Ser Asp Ser Asp Leu Phe Asp Asn Phe 105 Val Asn Phe Asp Ser Leu Lys Val Tyr Leu Asp Asp Trp Lys Asp Val Leu Pro Gln Gly Ile Ser Ser Phe Ile Asp Asp Ile Gln Ala Gly Asn 135 Tyr Ser Thr Ser Ser Leu Asp Asp Leu Ser Glu Asn Phe Ala Val Gly 155 Lys Gln Leu Leu Arg Asp Tyr Asn Ile Glu Ala Lys His Pro Val Val 170 Met Val Pro Gly Val Ile Ser Thr Gly\Ile Glu Ser Trp Gly Val Ile 185 Gly Asp Asp Glu Cys Asp Ser Ser Ala His Phe Arg Lys Arg Leu Trp 200 Gly Ser Phe Tyr Met Leu Arg Thr Met Val Met Asp Lys Val Cys Trp Leu Lys His Val Met Leu Asp Pro Glu Thr Gly Leu Asp Pro Pro Asn Phe Thr Leu Arg Ala Ala Gln Gly Phe Glu Ser Thr Asp Tyr Phe Ile 250 Ala Gly Tyr Trp Ile Trp Asn Lys Val Phe Gln\ Asn Leu Gly Val Ile 265 Gly Tyr Glu Pro Asn Lys Met Thr Ser Ala Ala Tyr Asp Trp Arg Leu Ala Tyr Leu Asp Leu Glu Arg Arg Asp Arg Tyr Phe Thr Lys Leu Lys Glu Gln Ile Glu Leu Phe His Gln Leu Ser Gly Glu Lys Val Cys Leu Ile Gly His Ser Met Gly Ser Gln Ile Ile Phe Tyr Phe Met Lys Trp Val Glu Ala Glu Gly Pro Leu Tyr Gly Asn Gly Gly Arg\Gly Trp Val Asn Glu His Ile Asp Ser Phe Ile Asn Ala Ala Gly Thr Leu Leu Gly Ala Pro Lys Ala Val Pro Ala Leu Ile Ser Gly Glu Met Ly's Asp Thr Ile Gln Leu Asn Thr Leu Ala Met Tyr Gly Leu Glu Lys Phe \Phe Ser

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Arg Ile Glu Arg Val Lys Met Leu Gln Thr Trp Gly Gly Ile Pro Ser
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Ser Glu Asp Ala Leu Asn Asn Asn Thr Asp Thr Tyr Gly Asn Phe Ile
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Lys Asp Ala Ile Asn Met Thr Leu Aer Ile Ser Pro Glu Trp Leu Gln
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Glu Val Pro Leu Pro Glu Ala Pro His Met Lys Ile Tyr Cys Ile Tyr
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Gly Val Asn Asn Pro Thr Glu Arg Ala Tyr Val Tyr Lys Glu Glu Asp
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Asp Ser Ser Ala Leu Asn Leu Thr Ile Asp Trr Glu Ser Lys Gln Pro
Val Phe Leu Thr Glu Gly Asp Gly Thr Val Pro\ Leu Val Ala His Ser
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Met Cys His Lys Trp Ala Gln Gly Ala Ser Pro Tyr Asn Pro Ala Gly
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Ile Asn Val Thr Ile Val Glu Met Lys His Gln Prb Asp Arg Phe Asp
Ile Arg Gly Gly Ala Lys Ser Ala Glu His Val Asp\Ile Leu Gly Ser
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320

Comp

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Ala Lys Tyr Ile Lys Ala Val Met Asn Ile Gly Gly Pro Phe Leu Gly
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Chr

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Glu Arg Ala Tyr Val Tyr Lys Leu Asn Gln Ser Pro Asp Ser Cys Ile
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Pro Phe Gln Ile Phe Thr Ser Ala\His Glu Glu Asp Glu Asp Ser Cys
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Leu Lys Ala Gly Val Tyr Asn Val Asp Gly Asp Glu Thr Val Pro Val
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Leu Ser Ala Gly Tyr Met Cys Ala Lys Ala Trp Arg Gly Lys Thr Arg
Phe Asn Pro Ser Gly Ile Lys Thr Tyr\Ile Arg Glu Tyr Asn His Ser
Pro Pro Ala Asn Leu Leu Glu Gly Arg Gly Thr Gln Ser Gly Ala His
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Val Asp Ile Met Gly Asn Phe Ala Leu Ile Glu Asp Ile Met Arg Val
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Asp Glu Asn Asn Lys Gly Gly Ser Val His Asn Lys Ard Glu Ser Arg
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96

CM

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							t ta Let									288
							agc Ser									336
							gtg Val 120									384
							ttt Phe									432
							gat Asp									480
							aat Asn									528
							acg Thr									576
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							aca Thr									672
							cct Pro									720
							ggc Gly									768
							aaa Lys									816

CI Cent

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						aga Arg 295										912
						cat His										960
att Ile	gga Gly	cat His	tct Ser	atg Met 325	ggt Gly	tct Ser	cag Gln	att Ile	atc Ile 330	ttt Phe	tac Tyr	ttt Phe	atg Met	aaa Lys 335	tgg Trp	1008
gtc Val	gag Glu	gct Ala	gaa Glu 340	ggc Gly	cct Pro	ctt Leu	tac Tyr	ggt Gly 345	aat Asn	ggt Gly	ggt Gly	cgt Arg	ggc Gly 350	tgg Trp	gtt Val	1056
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						gct Ala 375										1152
						gcc Ala										1200
						atg Met										1248
						gag Glu										1296
						aac Asn										1344
						agc Ser 455										1392
						aca Thr										1440
						tac Tyr										1488
						cta Leu										1536

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gta ttc ctc Val Phe Leu										1728
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ata cgt ggt Ile Arg Gly 610			Ala G			p Ile				1872
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Asn His Ile		Gln Glr	Gly L 40	eu Gly	His Ly	s Arg 45	Arg	Arg	Gly	
Ile Ser Gly 50	Ser Ala	Lys Arc		lu Arg	Gly Ly:	_	Phe	Asp	Arg	

CM

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aaa aga gac ggg aa Lys Arg Asp Gly As 65			
att ttc att ctt gg Ile Phe Ile Leu G			
ggc gct tat cat gr Gly Ala Tyr His Va 100	al His Asn Ser A	gat agd gac ttg t Asp Ser Asp Leu P 105	tt gac aac ttt 336 he Asp Asn Phe 110
gta aat ttt gat to Val Asn Phe Asp Se 115		Tyr Leu Asp Asp T	
ctc cca caa ggt at Leu Pro Gln Gly I 130			
tac tcc aca tct to Tyr Ser Thr Ser Se 145			
aaa caa ctc tta co Lys Gln Leu Leu An 16			
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						aac Asn										816
						atg Met										864
						aga Arg 295										912
						cat His										960
						tct Ser										1008
						ctt Leu										1056
						ttc Phe										1104
						gct Ala 375										1152
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						atg Met										1248
						gag Glu										1296
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Con

						-			J.	•						
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aaa Lys 465	gac Asp	gcc Ala	att Ile	aac Asn	atg Met 470	aca Thr	tta Leu	tcg Ser	ata Ile	tca Ser 475	cct Pro	gaa Glu	tgg Trp	ctc Leu	caa Gln 480	1440
						tac Tyr										1488
						cta Leu										1536
						gct Ala										1584
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						ttg Leu										1680
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ctc Leu	gtc Val	gag Glu	cca Pro	cgc Arg 645	caa Gln	ttg Leu	tct Ser	aat Asn	ttg Leu 650	agc Ser	çag Gln	tgg Trp	gtt Val	tct Ser 655	cag Gln	1968
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Chr

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35 40\Leu Gly His Lys Arg Arg Arg Gly

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Lys Arg Asp Gly Asn Gly Arg Lys Arg Trp Arg Asp Ser Arg Arg Leu
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Gly Ala Tyr His Val His Asn Ser Asp Ser Asp Leu Phe Asp Asn Phe
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Val Asn Phe Asp Ser Leu Lys Val Tyr Leu Asp Asp Trp Lys Asp Val
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Tyr Ser Thr Ser Ser Leu Asp Asp Leu Ser Glu Asn Phe Ala Val Gly
145 150 155 160

Lys Gln Leu Leu Arg Asp Tyr Asn Ile Glu Ala\Lys His Pro Val Val
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Met Val Pro Gly Val Ile Ser Thr Gly Ile Glu Ser Trp Gly Val Ile
180 185 190

Gly Asp Asp Glu Cys Asp Ser Ser Ala His Phe Art Lys Arg Leu Trp
195 200 205

Gly Ser Phe Tyr Met Leu Arg Thr Met Val Met Asp Lys Val Cys Trp 210 215 220

Leu Lys His Val Met Leu Asp Pro Glu Thr Gly Leu Asp Pro Pro Asn 225 230 235 240

Phe Thr Leu Arg Ala Ala Gln Gly Phe Glu Ser Thr Asp Tyr Phe Ile 245 250 255

Ala Gly Tyr Trp Ile Trp Asn Lys Val Phe Gln Asn Leu Gly Val Ile
260 265 270

Gly Tyr Glu Pro Asn Lys Met Thr Ser Ala Ala Tyr Asp Trop Arg Leu 275 280 285

Chr

Ala Tyr Leu Asp Leu Glu Arg\Arg Asp Arg Tyr Phe Thr Lys Leu Lys 295 Glu Gln Ile Glu Leu Phe His dln Leu Ser Gly Glu Lys Val Cys Leu 310 Ile Gly His Ser Met Gly Ser Glh Ile Ile Phe Tyr Phe Met Lys Trp Val Glu Ala Glu Gly Pro Leu Tyr\Gly Asn Gly Gly Arg Gly Trp Val Asn Glu His Ile Asp Ser Phe Ile Asn Ala Ala Gly Thr Leu Leu Gly 360 Ala Pro Lys Ala Val Pro Ala Leu Ile Ser Gly Glu Met Lys Asp Thr 375 Ile Gln Leu Asn Thr Leu Ala Met Ty $\eta$  Gly Leu Glu Lys Phe Phe Ser Arg Ile Glu Arg Val Lys Met Leu Gln Thr Trp Gly Gly Ile Pro Ser Met Leu Pro Lys Gly Glu Glu Val Ile Trp Gly Asp Met Lys Ser Ser Ser Glu Asp Ala Leu Asn Asn Asn Thr Asp Thr Tyr Gly Asn Phe Ile Arg Phe Glu Arg Asn Thr Ser Asp Ala Phe Asn Lys Asn Leu Thr Met Lys Asp Ala Ile Asn Met Thr Leu Ser Ile Ser Pro Glu Trp Leu Gln Arg Arg Val His Glu Gln Tyr Ser Phe Gly Tyr Ser Lys Asn Glu Glu 490 Glu Leu Arg Lys Asn Glu Leu His His Lys Hit Trp Ser Asn Pro Met 505 Glu Val Pro Leu Pro Glu Ala Pro His Met Lys\Ile Tyr Cys Ile Tyr Gly Val Asn Asn Pro Thr Glu Arg Ala Tyr Val Tyr Lys Glu Glu Asp 535 Asp Ser Ser Ala Leu Asn Leu Thr Ile Asp Tyr Gl↓ Ser Lys Gln Pro 550 555 Val Phe Leu Thr Glu Gly Asp Gly Thr Val Pro Leu\Val Ala His Ser 565 Met Cys His Lys Trp Ala Gln Gly Ala Ser Pro Tyr Asn Pro Ala Gly 585 Ile Asn Val Thr Ile Val Glu Met Lys His Gln Pro Asp Arg Phe Asp 595 600

Cl

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